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# Fusarium Blight of Soybeans

John Dunleavy
*U. S. Department of Agriculture*

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JOHN DUNLEAVY[2]

*Abstract. Fusarium orthoceras* produced necrosis of succulent root tissues of soybean seedlings and infected tips of lateral roots of older plants. Pods and seeds were most susceptible to fungus penetration after maturity. High relative humidity was necessary for infection of seeds. Maximum stand reduction and yield loss under field conditions were obtained when seeds were sown in Fusarium-contaminated soil before rains. In a greenhouse the disease was most destructive when plants were grown in soil at 100-percent water-holding capacity at 21° C. The disease was less damaging to plants grown in soil at the same moisture level at 27°. Seedlings grown in contaminated soil at 100-percent water-holding capacity at 21° for three weeks wilted permanently in a few hours when the temperature was raised to 33°, whereas plants in noncontaminated soil did not wilt.

Fusarium species have been reported to be pathogenic on soybeans, *Glycine max* (L.) Merr., by Cromwell (1917), Armstrong and Armstrong (1950, 1958), Liu (1940), Nojima (1926), Dunleavy (1954, 1955, 1956), and Neeley (1957). Gerdeman (1954) found several Fusarium species associated with a root rot of soybeans but believed they were saprophytes or secondary pathogens. Most investigators experienced difficulty in obtaining consistent infection and a few could not obtain infection except under special conditions.

A disease caused by a Fusarium species has been observed in Iowa since 1953. Some disease symptoms were similar to those described previously; others were different. Although the disease was not as destructive to older plants as some pathogens of soybeans, it is important because of the general debilitating effect the fungus has on seedlings and young plants. This study was conducted to obtain additional information concerning Fusarium blight and, particularly, to investigate the factors affecting plant infection.

## MATERIALS AND METHODS

Fusarium isolates were obtained from diseased soybean seedlings and subcultured. Isolates were maintained by transferring a mass of mycelium and conidia to fresh medium. A single typical isolate was used for preparation of inoculum for all experiments.

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[2] Plant Pathologist, Crops Research Division, U. S. Department of Agriculture, and Iowa Agricultural and Home Economics Experiment Station, Ames, Iowa.

Inoculum for greenhouse tests was prepared by washing conidia from ten two-week-old cultures in Petri dishes with sterile distilled water and diluting to 100 ml. Ten ml of inoculum were added to each four-inch pot of soil and ten seeds were sown immediately. All treatments in greenhouse tests with the exception of one in which wooden boxes were used, were replicated six times (six pots per treatment). Plants for one experiment were grown in soil in wooden boxes 1 foot square and 4 feet high. Fifty ml of inoculum were used and ten seeds were sown in each box. Sterile distilled water was added to control pots or boxes of inoculum.

Soil moisture level was expressed as percentage of maximum waterholding capacity of the soil (field capacity) and was maintained by weighing pots of soil each day. Sufficient water was added daily to maintain the various moisture levels.

Inoculum for field experiments was prepared by growing the fungus on a grain mixture composed of equal parts of moist, sterile corn and oats. One pint of inoculum was distributed to each 20-foot, open furrow about 2 inches deep. Inoculum was covered with soil and 200 seeds planted immediately. Stand counts were recorded 5 to 6 weeks after seeds were sown. Rows were trimmed to 16 feet before harvest.

Maneb fungicide was applied in a spray containing a wetting agent at the rate of three pounds per acre.

Unless otherwise stated, Hawkeye soybeans were used.

## Results

*Field symptoms.* The first evidence of the disease in the field was poor germination in sections of rows. Plants emerged late in some of these and were stunted. The root systems of severely infected plans were sometimes completely destroyed (Fig. 1,A). Destruction of root tissue usually stopped near the zone between root and stem, and if the seeds had been planted deep enough, lateral roots usually developed from this zone. Such plants continued to develop but were stunted. Infected cotyledons (Fig. 1,*C*) first became chlorotic and then necrotic. The cortex and root tips of healthy-looking plants grown in contaminated soil were infected. Infection stimulated production of new lateral roots from the older portions of infected roots. Considerable infection of this type resulted in a shallow, fibrous root system lacking a tap root.

Only rarely were plants beyond the seedling stage killed. The only symptom expressed by the above-ground portion of plants

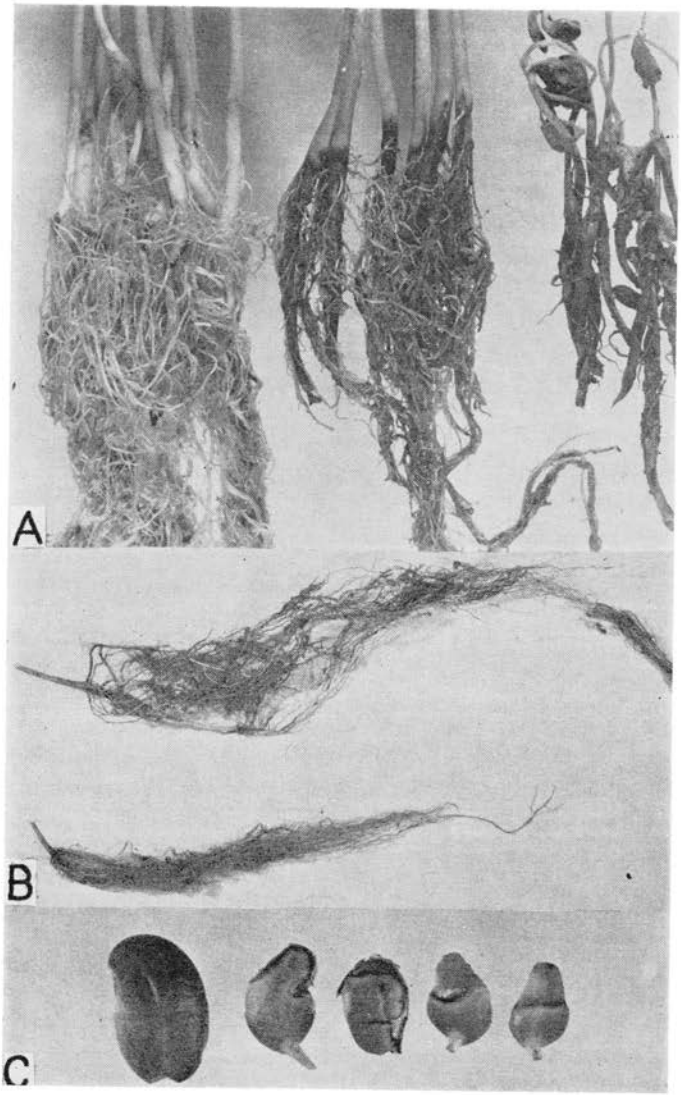


Figure 1. A, left to right, roots of healthy soybean seedlings, roots infected with *Fusarium orthoceras*, roots of seedlings killed by the fungus. B, top to bottom; healthy, mature soybean root system and a mature Fusarium-infected root system. C, healthy soybean cotyledon (left) and Fusarium-infected cotyledons in various stages of decomposition.

in most cases was slight to moderate stunting. The wilt symptoms described by Cromwell (1917), Neeley (1957), and Armstrong and Armstrong (1958) were never observed on plants over six weeks old.

The spring of 1959 was exceptionally wet. Counties in south-

central Iowa received up to 10 inches of rain in May. Stands were not as good as normal, and Fusarium blight was reported from many areas. After the very wet May, five counties in the affected area received little or no rain during the first three weeks of June. Plants were cultivated, the upper two or three inches of soil dried, and consequently the lower leaves wilted permanently. These plants would have died during the following two weeks without additional moisture, but healthy plants developed normally in the same soil. Wilting ceased and plant development continued when rains occurred late in June.

*Pod infection.* Pod infection was difficult to detect in the field. It was most frequently observed during prolonged rainy periods or in especially damp locations late in the growing season as plants approached maturity. Infected pods or seeds were detected by examination under the microscope for mycelial growth and spore production. Infected areas could be more readily ascertained by placing pods or seeds in a Petri dish containing 5 to 10 ml of water for 24 to 48 hours. Heavy mycelium developed over infected areas. Little or no growth occurred in healthy pods or seeds.

During a cloudy period nearly mature plants with yellow pods were inoculated with a suspension of conidia and covered with a plastic sheet for 48 hours. Plants in adjacent rows were sprayed with maneb and also covered. Each treatment was replicated four times. After one week 100 seeds were removed from pods in each treated row and placed in Petri dishes containing water. The percentage of infected seeds was determined after 48 hours. Mean percentage of seeds infected on inoculated plants was 24 percent as compared with 8 percent on the noninoculated plants, indicating that some natural infection had occurred before inoculation.

Comparative susceptibility of pods at various stages of development was determined. Plants with pods in various stages of development were sprayed with a suspension of conidia and placed in a moist chamber for four days. They were then removed and pods allowed to mature (Table 1). Maximum infection was obtained when mature pods were inoculated. Infected seeds could not be distingushed from healthy ones after removal from pods.

*Root infection.* Numerous observations indicated that severe disease usually followed when contaminated soil was saturated with water soon after seeds were sown. An entire soybean research nursery was destroyed by the disease when 3½ inchs of rain fell shortly after planting. Only pure cultures of *Fusarium orthoceras* Appel & Wr. were obtained. Other soybean pathogens

Table 1. Percentage infection of soybean seeds[a] from pods inoculated with *Fusarium orthoceras* at various stages of development.

| Stage of pod development | Replication 1 | 2 | 3 | 4 | Mean |
|---|---|---|---|---|---|
| 1 in. long, flat, seeds developed | 0 | 0 | 0 | 0 | .. |
| 1½ in. long, pod wall distended, seeds half developed | 0 | 0 | 0 | 0 | .. |
| 1¾ in. long, pod walls fully distended, green | 20 | 4 | 4 | 12 | 10 |
| Pods yellow | 52 | 36 | 28 | 44 | 40 |
| Pods mature | 76 | 64 | 52 | 48 | 60 |
| Control, pods mature, noninoculated | 0 | 0 | 0 | 0 | .. |

[a] Based on a sample of 25 seeds

such as *Pythium*, *Rhizoctonia*, and *Phytophthora* were absent although special techniques were used in attempts to isolate them.

In another case two replicates of an experiment plot involving *F. orthoceras* were planted before a 2-inch rain stopped the work. The remaining two replicates were planted four days later. Plant stands and yields from seeds sown in Fusarium-contaminated soil before the rain were very poor, but stands and yields from plantings made after the rain were moderately good (Table 2). Prior experiments had given essentially the same results as those from the planting made after the rain. Stands were usually poor, but yields were affected only slightly or not at all.

Considering all varieties, the stand from the planting made on contaminated soil before the rain was reduced 64 percent as compared with an 11 percent reduction on the noncontaminated soil. Plants from seeds sown in contaminated soil before the rain

Table 2. Mean stands and yields of soybean plants grown in soil contaminated with *Fusarium orthoceras* and in noncontaminated soil. Half the seeds were sown immediately before a 2-inch rain and the other half 4 days later.

| Variety | Mean percentage stand[a] Before rain C[b] | Before rain NC[c] | After rain C | After rain NC | Mean yield (bu/a) Before rain C | Before rain NC | After rain C | After rain NC |
|---|---|---|---|---|---|---|---|---|
| Earlyana | 7 | 66 | 22 | 76 | 10 | 25 | 31 | 34 |
| Blackhawk | 6 | 82 | 70 | 84 | 8 | 27 | 29 | 33 |
| Richland | 14 | 80 | 85 | 99 | 22 | 32 | 38 | 36 |
| Hawkeye | 8 | 80 | 80 | 93 | 16 | 35 | 36 | 42 |
| Mean | 9 | 77 | 64 | 88 | 14 | 30 | 34 | 36 |

[a] Based on 200 seeds
[b] Contaminated soil
[c] Noncontaminated soil

yielded 59 percent less than plants from seeds sown later, whereas the yield reduction was only 17 percent on noncontaminated soil. Presence of some *F. orthoceras* in the noncontaminated field soil may account for the reduction in stand and yield observed on this soil. A comparison of stand and yield on contaminated soil before the rain with that on noncontaminated soil after the rain showed that stand was reduced 79 percent and yield 61 percent.

Table 3. Comparison of soybean plants grown in soil contaminated with *Fusarium orthoceras* and control plants grown in noncontaminated soil.

| Observation | Contaminated soil | | | Control |
|---|---|---|---|---|
| | Box 1 | Box 2 | Box 3 | |
| Days till emergence | 12 | 11 | 11 | 8 |
| Stand (before thinning) | 10 | 9 | 8 | 10 |
| Mean number of pods per plant (3 plants per box) | 72 | 83 | 84 | 170 |
| Mean yield per plant (gms) | 14 | 20 | 17 | 50 |
| Mean dry weight of plant roots (gms) | 9 | 8 | 6 | 9 |
| Mean dry weight of plant stems (gms) | 21 | 18 | 13 | 41 |
| Mean weight of 100 seeds (gms) | 10 | 12 | 9 | 15 |

Soybean plants grown in soil contaminated with *F. orthoceras* were compared with control plants grown in noncontaminated soil in large wooden boxes in the greenhouse. Plants in contaminated soil emerged three or four days later than control plants and stands were slightly lower (Table 3). There were about half as many pods per plant on diseased plants as on control plants and yields were reduced over 40 percent. There was little difference in weight of roots from diseased and healthy plants, but the mean dry weight of stems from diseased plants was approximately 50 percent that of the control plants. Seeds of diseased plants were smaller and less dense than those from control plants. Pods and leaves of diseased plants were also smaller. Control plants were slightly taller than diseased ones and matured about two weeks later.

Plant roots were carefully washed from the soil to obtain the entire root system. Roots from diseased plants were very small and fibrous, whereas roots of healthy plants had very well-developed tap roots and comparatively few fibrous roots (Fig. 1,*B*). Tap roots had been rotted from most diseased plants as seedlings. Fibrous roots developed from the unrotted, upper portion of the tap root and adventitiously from the base of the stem.

The effects of soil moisture and temperature on disease de-

velopment were determined in another greenhouse experiment. Seeds were sown in Fusarium-contaminatd soil and in noncontaminated soil that ranged from 40 to 100 percent of field capacity. Plants were grown at 21° C and 27° C.

The disease was most severe when the moisture level was maintained at 100 percent of field capacity at 21° C (Table 4). Plants grown in contaminated soil were only half as tall as those grown in noncontaminated soil. Differences in height of plants grown in contaminated and noncontaminated soil at the different moisture levels were not as pronounced at 27° as at 21°. At 40 percent of field capacity effects of the disease were more pronounced at 27° and less pronounced at 21°. Stands were approximately 20 to 30 percent lower in contaminated soil at all soil moisture levels and at both temperatures.

Table 4. Mean plant height and percentage stand obtained when soybean seeds were planted in soil contaminated with *Fusarium orthoceras* and in noncontaminated soil of different moisture levels.

| Soil moisture (percentage of field capacity) | 21° C | | | | 27° C | | | |
|---|---|---|---|---|---|---|---|---|
| | Mean plant height (cm) | | Mean[a] percentage stand | | Mean plant height (cm) | | Mean percentage stand | |
| | C[b] | NC[c] | C | NC | C | NC | C | NC |
| 40 | 18 | 22 | 66 | 96 | 15 | 20 | 62 | 94 |
| 60 | 16 | 23 | 63 | 90 | 16 | 21 | 65 | 94 |
| 80 | 14 | 24 | 68 | 90 | 18 | 22 | 70 | 93 |
| 100 | 12 | 24 | 63 | 95 | 22 | 24 | 74 | 96 |

[a] Based on 60 plants
[b] Contaminated soil
[c] Noncontaminated soil

Because plant water loss through transpiration was probably greater at 27° C than at 21°, it was believed disease effects observed on plants grown in contaminated soil at 40 percent of field capacity at 27° were due to a combination of disease and insufficient water. This hypothesis was tested in an additional experiment using twice the usual number of pots of contaminated and noncontaminated soil. Seeds were sown in soil at 100 percent of field capacity and the plants grown at 21° for 3 weeks. Half the pots were then placed in another greenhouse at 33°. Plants growing in contaminated soil at 33° were permanently wilted in a few hours, whereas plants in noncontaminated soil did not wilt. None of the plants grown at 21° wilted.

## Discussion

Lack of consistent results with the usual inoculation procedures led some earlier workers to conclude that certain Fusarium species were nonpathogenic on soybeans or of little consequence in causing disease. Armstrong (1941) and Neeley (1957) obtained consistent results by inoculating roots of plants grown in a nutrient solution. Neeley also reported production of

a toxin by *F. orthoceras* in liquid medium that caused severe symptoms on soybeans.

One interpretation of the data presented here is that *F. orthoceras* produced more toxin in soil saturated with water at 21° C than in unsaturated soil, and that more toxin was produced at 21° than at 27°. Even though the disease was less severe on plants grown in saturated soil at 27°, it produced stunting in soil of medium or low moisture level because of the increased transpiration rate.

Postulation of a fungus-produced toxin also fits the observed symptoms. Succulent root parenchyma, cortical tissue of seedling roots, and root tips of older plants were most seriously infected. Root tissue was observed to be discolored far in advance of hyphal penetration of the fungus. Secondary root tissues were not as seriously infected. The fungus penetrated lignified or corky tissue only rarely.

Field observations and data indicate the disease is widespread in Iowa and Missouri. Under average growing conditions the disease may result in a reduced stand and slight to moderate general depression of growth. Heavy rains and cool temperatures after planting in contaminated soil, or after planting infected seed, are likely to result in reduction of stand and considerable yield loss. High temperatures and low soil moisture levels following seedling infection tend to aggravate the diseased condition of the plants through depression of growth and can cause death of such plants when healthy plants are thriving.

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